STUDIES ON SERUM ZINCEMIA IN 202 PERSONS WITH POSTHEPATITIC SYNDROMES AND AFFECTIONS

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Zinc is an element playing an important role in intermediate metabolism, owing to its relationship to the active center of a number of ferments (2). This was demonstrated in a clearcut manner after Keilin and Mann (1944) succeeded in isolating carbonic anhydrase in 1944 and established that it contains 0.33% zinc, and after Vallee and co-workers discovered zinc in some dehydrogenases, peptidases, transphosphorilases and the like (32, 33). Similarly, it has been long since pointed out that distribution of zinc in the various organs is regulated by the liver (Shelline, Chaikoff and assoc. 1943, cited by 25). Ever since then, intense studies on the zinc metabolism in various hepatic diseases was begun. It has been found out that virus hepatitis (vh) runs a course with transitory lowering of zinc concentration in the whole blood (1) and serum (3, 18, 25, 38). Any way, there are reports on investigations by other authors in which a similar reduction has not been established (4). In the urine of vh patients, an increased (1), or normal zinc content (3) has been found. The zinc level in the liver of vh patients is reduced, some times up to 2—2.5 times that of the normal (22). During the healing stage, the normalization of zinc in the blood and urine lags behind the normalization of some of the laboratory indices (1). The slow normalization of the serum zinc is explained by the deep alteration of some of the liver functions. It was established that in alcoholic and posthepatitis cirrhosis the zinc concentration in the serum decreases (4, 24, 25, 26, 36, 37, 38), whereas the zinc amount eliminated via 24-hour urine is increased, sometimes quite significantly (4, 24, 27, 36). The zinc content in the cirrhotic liver is low (10, 13, 15, 20, 21, 22, 24, 25, 26, 29, 34, 35, 36 and others). Vallee and co-workers established that there is a correlation between the clinical picture of cirrhosis, resp. its early or advanced phase, and the decrease of zinc in the serum, resp. its rise in the urine (29, 34, 35, 36). The hepatic coma is accompanied by very strong reduction of the serum zinc (according to 4).

Based on the facts referred to above, we investigated the serum concentration of zinc in subjects with past illness of virus hepatitis and compared the results with the subjective and objective symptoms and with the clinico-laboratory indices of those investigated. The purpose was to see if in the various groups of the total number of affected with virus hepatitis in the past (e. g. cured, with chronification of the process, cirrhoses) there are changes in the serum zinc, and up to what extent would they be considered as an additional index in evaluating the curing, respectively chronification of the condition. The present report is a continuation of earlier investigations by the same authors (5, 6).

Material and Method

A series of 202 persons with past history of virus hepatitis, dating back 6 months to 20 years, were investigated. The age of the patients ranged from 5 to 78 years, with a net predomination of the cases in the 25—50 years age group. The males were 100 and the females — 102. The investigations were made on the basis of 26 subjective, 15 objective (physical) and 33 clinico-laboratory indices. According to the results recovered, the total number was divided into 6 groups, in accordance to the overall assessment of the liver after the affection:

1) *Cured* — 92 cases
2) *Not cured* (i. e. with protracted process) — 47 cases.
3) *Isolated residual hepatomegaly* («fibrosis», «chronic inactive hepatitis», without evidence for activity of the process) — 22 cases.
4) *Posthepatitis hyperbilirubinemia (PHHB)* — 5 cases.
5) *Chronic hepatitis* — 27 cases.
6) *Posthepatitis cirrhosis* — 9 cases.

Biopsy diagnosis was established in isolated cases only.

The serum zinc was demonstrated according to the dityson method of Rechenberger. The mean zincemia established in 22 healthy persons amounted to 132±23 gamma % at M=4.9. We assumed that zincemia decrease was present whenever its concentration was beneath 109 gamma %.

Results. The overall results of the investigation of zinc in the series of 202 cases with past history of virus hepatitis, distributed in the 6 groups, are the following: (Table 1).

Table 1

Results of zinc level investigations in 202 individuals with past virus hepatitis illness

Group	Cases	Reduced zinc	Normal zinc	Mean zinc concentration γ %
1. Cured	92	25	67	126.22
2. Uncured	47	17	30	125.85
3. Residual Hepatomegalia	22	9	13	128.22
4. Posthepatitis hyperbil.	5	2	3	115.22
5. Chronic hepatitis	27	21	6	104.70
6. Posthepatitis cirrhosis	9	8	1	71.00

The lowest value substantially beneath the normal zincemia — 71 gamma % — is noted in cirrhosis. Next, similarly with low mean zincemia — 104 gamma % — is the group of chronic hepatitis. The above figures are statistically reliable as compared to the group of cured, as well as between each other. In the remaining 4 groups (1, 2, 3 and 4), the mean zinc concentration is within normal limits, and in isolated cases — above the norm. Groups 2, 3 and 4 did not show statistically reliable zinc reduction as related to group 1 — the cured, and between each other neither.

Group 1. We assumed as cured the cases which, after the vh illness, were free of subjective and objective signs for hepatic disorders and revealed

normal clinico-laboratory indices of the condition of the liver. The mean serum zincemia in the group, comprising 92 investigated subjects, was within the normal limits — 126.22 gamma %. In 25 cases the zincemia was between 87 and 106 gamma % (27.1%), and in the remainder (67) — the zincemia was normal or slightly exceeding the norm. The time elapsed after the acute affection in 25 cases with reduced zinc was between 0.5 and 11 years, and in the 67 cases with normal zinc — between 0.5 and 13 years.

Group 2. We assumed as UNCURED acute hepatitis cases those which after the sixth month and before the second year from the acute stage of the disease, have usually insignificant subjective complaints, without or with but slight hepatomegaly, and with one to three clinico-laboratory indices for hepatic damage (urobilinogenuria, hypergammaglobulinemia, increased Weltmann's column or pathological MacLagan test). The mean serum zincemia of the 47 subjects investigated from this group was normal — 125 gamma %. Decrease of zinc was recorded in 17 cases (36.1%), the concentration ranging from 70 to 106 gamma %. In the remainder (30 cases), the zincemia ranged between 110 and 225 gamma %. No dependence was established between the degree of zincemia and the results of the routine indices for hepatic lesion.

Group 3. It includes the cases in which a certain, more frequently insignificant hepatomegaly is established after the second year from the onset, without or in single cases, with insignificant symptomatics (usually slight pains or heaviness in the region of the liver following physical strain), and in most of the patients, with lack of even one pathological, clinico-laboratory index. Out of 22 subjects investigated, a decreased zinc was noted in 9 (40.9%), with zinc fluctuations from 74 to 106 gamma %. Normal zinc was recorded in 13 cases (between 115 and 225 gamma %). The mean zincemia for the group amounted to 128.22 gamma %.

Group 4. Out of the five persons investigated with **posthepatitis hyperbilirubinemia,** reduced zinc values were established in two cases (76 and 106 gamma %).

Group 5. The group of chronified hepatitis comprised 27 subjects. Their vh illness dated back more than 2 years ago. All exhibited pronounced subjective symptomatics, hepato-, spleno- or hepatosplenomegaly, plus more than three pathological results of the laboratory indices, considered most important for chronic hepatic damage (urobilinogenuria, hypergammaglobulinemia, hyperbilirubinemia, hypoalbuminemia, positive flocculation reactions). The serum zinc was found to be reduced in 21 of the 27 persons investigated (77.7%). The zincemia concentration ranged from 60 to 108 gamma %.

We failed to establish an interdependence between zincemia, on the one hand, and duration of the disease, subjective complaints, objective symptoms and laboratory tests, on the other.

Six of the patients disclosed normal zincemia — from 135 to 177 gamma%. These patients did not differ significantly from the other 21 cases with reduced zinc, neither in terms of duration of the affection, nor in terms of symptomatology presented. The most frequently met with clinical and clinico-laboratory indices in the group of 27 patients are illustrated in Table 2.

Table 2

The most frequently met clinical and clinico-laboratory indices in 27 patients with chronic hepatitis

Indices	Cases
1. Subjective complaints	27
2. Weltmann reaction above 7 test-tubes	25
3. Hepatomegalia	24
4. Hyperurobilinogenuria	23
5. **Hypozincemia**	21
6. MacLagan reaction above 40 PhU	18
7. Hypergammaglobulinemia (above 23%)	16
8. Hyperbilirubinemia	12
9. Icterus on the skin or sclera	9
10. Splenomegalia	7
11. Anemia (Hb beneath 70%)	6
12. Increased transaminases	5
13. Leukopenia (leuk. beneath 5000)	5
14. Thrombopenia (thrombocytes beneath 200000)	4
15. Telangiectasis	4
16. Emaciation	4 etc.

It is evident from the table that in chronic hepatitis, the hypozincemia is inferior in terms of frequency of the subjective symptoms, increased Weltmann's column, hepatomegaly and hyperurobilinogenuria, and exceeds in terms of incidence of such important indices as the MacLagan reaction, hypergammaglobulinemia, splenomegaly and hyperbilirubinemia.

Table 3

Dependence between Zincemia and Gammaglobulinemia in Cases with Posthepatitis Cirrhosis

Cases	Gamma-globulins %	Zinc γ %
1. M. P. M.	43	25
2. T. M. K.	40	54
3. H. N. A.	36.3	86
4. M. D. M.	30.8	80
5. B. Y. M.	28	141
6. N. N. P.	27.5	58
7. Y. E. Z.	26.3	70
8. T. S. T.	24	80
9. S. D. S.	20	45

Group 6. It comprises 9 cases with posthepatitis cirrhosis, 8 of which in advanced stage (5 with ascites). The zinc was decreased in 8 cases (88.8%), exhibiting a fluctuation between 25 and 141 gamma %. The mean zincemia of the group amounted to 71 gamma %. The lowest zinc values — 25 and 45 gamma% — were recorded in patients with greatly advanced, ascitic stage, with hepato- and splenomegaly, large gynecomastia in one of them and length of the postacute stage of the disease ranging from 4 to 20 years. The only patient with normal zincemia (141 gamma %) concerned a girl with ascites and duration of vh 6 months.

No strict interdependence was established between the degree of zincemia and the other clinical and clinico-laboratory indices. A certain de-

gree of correlation was however discovered between hypozincemia and hypergammaglobulinemia (Table 3).

The incidence of hypozincemia with posthepatitis cirrhosis in our series came after the subjective complaints, hyperurobilinogenuria, increased

Table 4

The most frequently met clinical and clinico-laboratory indices in 9 patients with posthepatitis cirrhosis

Indices	Cases
1. Subjective complaints	9
2. Hyperurobilinogenuria	9
3. Weltmann reaction above 7 test-tubes	9
4. Hyperbilirubinemia	9
5. **Hypozincemia**	8
6. MacLagan reaction above 40 PhU	8
7. Hypergammaglobulinemia	8
8. Hypoalbuminemia (alb. beneath 55%)	7
9. Hepatomegalia	7
10. Telangiectasis	7
11. Emaciation	7
12. Ascites	6
13. Anemia (Hb beneath 70%)	6
14. Palmar erythema	5
15. Splenomegalia	5
16. Leukopenia (leuk. beneath 5000)	4
17. Icterus on the skin or sclera	3 etc.

Weltmann's column and hyperbilirubinemia, recorded in the 9 cases. Hypozincemia was established as often as the pathological reaction of MacLagan and hypergammaglobulinemia — in 8 of the nine cases (Table 4).

Discussion and Inference

Among the group of persons with past history of virus hepatitis, there was not a single case with evidence for pancreatic, renal, infectious, neoplastic, blood or other diseases which are known to cause occasionally, a decrease of the serum zinc. Hence, the low values of zinc in our series should be explained by a process developing in the liver. We can't make a definitive statement that the 6 groups, differentiated in the present study, comprise only the diagnoses referred to the respective groups. Exceptions are by all means possible, since the biopsy of the liver has not been routinely and regularly done. Nevertheless, we have strived towards a long-term and critical re-evaluation of the diagnoses (secondary follow-up examinations of a great number of patients), on the basis of the overall clinical and laboratory finding concerning the hepatic state in each individual case.

From the data obtained, it is obvious that the cases with hypozincemia show a steady rise, to begin with the group of cured where they amount

to 27% and passing to the group of not cured — 36%, with residual hepatomegaly — 40%, chronic hepatitis — 77% and finally, reaching the group with posthepatitis cirrhosis, where the hypozincemia cases show the highest percentage — 88% (Fig. 1).

The second fact worth noticing is that the lower limit of the hypozincemic values reveals a progressive fall in the direction from group I to group VI — accordingly 87, 70, 74, 60, 60 and 25 gamma % (Fig. 1).

A third circumstance, although not so obvious, owing to the small number of observations, is that certain conformity was established between the degree of hypozincemia and the rise of gammaglobulin fraction in the serum, particularly in group VI (Table 3). In this respect, we would like to quote a similar finding made by Wolf (38), in the course of personal investigations.

It might be assumed that insofar as the progressive rise of hypergammaglobulinemia is considered as an important index about chronification and cirrhogenic processes in the liver, the steady reduction of the serum zincemia, in the course of months and years after the vh illness, is also (probably) one of the symptoms characteristic of these processes. In this respect we would add also the correlation established between hypozincemic values in these cases and the hyperurobilinogenuria and increased Weltmann's reaction etc in groups 5 and 6.

The finding of progressive reduction of the dynamically traced up zincemia is, in all likelihood, an additional indication that the process in the liver is not discontinued, but on the contrary, it persists and progresses with the eventual possibility of developing cirrhosis which is characterized by the lowest serum zinc concentrations known hitherto.

Our data demonstrate that the normalization of zincemia after vh constitutes a rather inert, slow process. The practical curing of hepatitis, although in a limited number of cases, might be associated, for some period of time, with a slightly reduced zincemia. Similar inferences have been reached by other authors also (3). In this respect, the sluggish dynamics of zincemia is in contrast to its prompt alteration observed in patients in the reconvelescent, postcoronary infarction period (7) and suggests the possibility for the existence of various mechanisms of changes.

The item of the essence of the mechanisms through which chronic hepatopathies lead to decrease of the zinc level in the serum and in the hepatic tissue is scarcely investigated and for the time being we do not avail of an acceptable explanation.

More investigations have been made on the influence of alcohol upon the zinc-metalloenzymes in the liver — alcohol dehydrogenase, glutamate dehydrigenase etc — as well as with the purpose to find an explanation of the paucity of zinc loss in the hepatic tissue in alcoholic cirrhosis (11, 26, 29, 30, 34). The chronic alcoholic intoxication causes a reduction of the activity of alcohol dehydrogenase in the liver (11, 26). It is possible that detachment of the zinc ion from the active center of the ferments is concerned, with intense discharge of zinc in the urine and fall of the serum and tissue zinc level.

Rechenberger and co-workers (24, 25) presume that the reduced zinc content in the serum in cirrhosis is a manifestation of systemic affection of the zinc metabolism, subsequent to the cirrhogenic lesion of the hepatic parenchyma, resulting in the occurrence of a negative zinc balance.

Others, doubtful of the alcohol effect upon zinc-metalloenzymes, assume that the rising dysproteinemia in the course of hepatopathies might cause a defect in the zinc-binding capacity of the plasmatic and tissue proteins, which in turn, might lead to disorder in the zinc balance of the organism (15, 18).

The studies in this respect are rendered rather difficult as the zinc in the blood is not bound to a specific protein carrier. Zinc fractions in the blood are known, carried by the albumins, alpha$_1$, alpha$_2$, beta$_1$, beta$_2$ and gamma globulins (9). Firmly bound zinc exists — about 34%, probably, as a zinc-proteid of alpha-1 and alpha-2 globulins, and labile bound zinc — about 66%, carried by the albumins and beta-globulins (4).

Reports on studies on the causes of the organism getting poor in zinc content subsequent to vh affection were not found in the pertinent literature. It is a well known fact that some of the zincmetalloenzymes during vh affection, exhibit higher activity in the blood, such as glutamate dehydrogenase, which normally is not found in the blood, lactate-lactic dehydrogenase and others.

In general outline, the metabolism of zinc in parenchymal lesions of the liver (hepatitis, cirrhosis) reveals certain characteristic features, distinguishing it from the metabolism of the other metals. For instance, copper in vh (8) and in cirrhosis is increased in the serum, and in cirrhosis — in the liver too, iron in vh is increased in the serum and lowered in the liver, and in cirrhosis it is reduced in the serum and increased in the liver, magnesium is decreased in the serum in alcoholism and cirrhosis (12, 19, 28), and increased in renal insufficiency (14) and so on. On the contrary, in acute and chronic hepatic parenchymal diseases, a total impoverishment of zinc is observed, involving the serum, hepatic tissue and sweat secretion (23), associated with concomitant increased urinary discharge rate.

The inferences reached on the ground of the results obtained are the following:

1. Persisting or progressive hypozincemia in subjects with past history of vh is an unfavourable symptom, indicating a probable tendency towards chronification of the process in the liver, provided all other causes for the lowering of the zinc level are ruled out.
2. The hypozincemia is a late sign, manifested at the time when other symptoms occur and characteristic of the advanced stage of the disease.
3. In subjects with cured vh, there might be some decrease of zinc in the serum, which should not be considered as a poor prognosis.
4. The hypozincemia in persons with past history of vh is an index of weak dynamics.

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ИЗУЧЕНИЕ СЫВОРОТОЧНОЙ ЦИНКЕМИИ У 202 ЛИЦ С ПОСТГЕПАТИТНЫМИ СИНДРОМАМИ И ЗАБОЛЕВАНИЯМИ

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РЕЗЮМЕ

Авторы исследовали сывороточную цинкемию у 202 лиц, переболевших вирусным гепатитом в период от 0,5 до 20 лет. Содержание цинка определялось по дитизонному методу Регенбергера. Снижение цинкемии допускается при величинах ниже 109%. Исследуемые были разделены на группы в зависимости от состояния печени.

Наиболее низкая, намного ниже нормальной, цинкемия — в среднем 79% была обнаружена у перенесших послегепатитные циррозы. На втором месте по снижению цинка является группа страдающих хроническим гепатитом — средняя величина 104%. Эти числа статистически достоверны в сравнении с группой выздоровевших (средняя цинкемия 126%). В остальных трех группах — невыздоровевшие от вирусного гепатита, с остаточной гепатомегалией и постгепатитной гипербиллирубинемией, средние концентрации цинка в границах нормы и статистически недостоверны.

Гипоцинкемия у переболевших вирусным гепатитом, имеющих поражения печени, является показателем, который по своей частоте превышает некоторые обычные показатели поражения печени, а именно: гипергамаглобулинемию, желтуху, спленомегалию, тимоловую пробу, гипербилирубинемию и пр. и уступает по частоте субъективным жалобам, удлиненной ленте Вельтмана, гиперуробилиногенурии и гепатомегалии. Цинкемия при поражении печени является сравнительно поздним симптомом.